

Four new solitary entoprocts (Entoprocta: Loxosomatidae) from Okinawa Island, The Ryukyu Archipelago, Japan

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Abstract.—Four new solitary entoprocts are described from Okinawa Island, the Ryukyu Archipelago, Japan. Based on the foot structures and the budding modes, three of them are assigned to the genus *Loxosomella* Mortensen, 1911 (*L. stomatophora*, new species; *L. lecythifera*, new species; and *L. intragemmata*, new species) and the other to the genus *Loxocorone* Iseto, 2002 (*L. dicotyledonis*, new species). They were found on glass slides or a glass bottle collected from a shallow reef flat, and thus, they are not commensal, unlike most of the other species hitherto described from other localities.

To date, a total of approximately 170 species are recognized in the phylum Entoprocta, of which about 120 species are solitary and the remainder are colonial. It is believed that a great number of unknown species are yet to be described for this phylum (Nielsen 1989, Wasson 2002), and thus, taxonomic study, describing new species, is still a principal subject for its biology and is essential to document its diversity. The solitary entoprocts (= the family Loxosomatidae Hincks, 1880) are currently classified into five genera based on foot structures and budding modes (Iseto 2002). Species of the genus *Loxosoma* Keferstein, 1863 have a disk-shaped foot, a sucking disc by which the animals can attach to and detach from substratum. Species of *Loxosomella* Mortensen, 1911 and *Loxocorone* Iseto, 2002 have, at least in the bud stage, an antero-posteriorly elongated foot with a foot gland and a foot groove that enables the animal to glide over the substratum. However, the two genera are distinguished by differences in their budding mode. The buds of the former genus attach to the body of the parent by the posterior tip of the foot, in contrast to the buds of the latter genus that attach to the parent by the back of the stalk or the calyx. The genus *Loxomitra*

Nielsen, 1964 resembles the genus *Loxocorone* in the budding mode; the buds attach to the parent by the back of the stalk. The buds of this genus, however, lack the foot groove and have a pair of small lateral expansion termed “terminal wings” (Nielsen 1989) at the tip of the stalk. The genus *Loxomespilon* Bobin & Prenant, 1953 is a monotypic genus; the sole component species of this genus, *L. perezii*, lacks a definite foot and stalk and attaches to the substratum, the body surface of the host polychaetes, by an attaching organ at the basal end of the calyx.

Solitary entoprocts have been reported from tropical areas to polar regions, from shallow to deep (> 500 m) waters. However, in most regions reports are still scarce, except in Europe where this animal group has been surveyed most intensively (Prenant & Bobin 1956, Nielsen 1989). Recently, three *Loxosomella* (*L. aloxiata*, *L. lappa*, *L. monocera*), two *Loxomitra* (*L. mizugamaensis*, *L. tetraorganon*), and one *Loxocorone* (*L. allax*) were described from coral reef shores of the Okinawa Islands, the middle part of the Ryukyu Archipelago, Japan (Iseto 2001, 2002). Since the entoproct fauna of this area has still not been studied sufficiently, extended surveys are expected

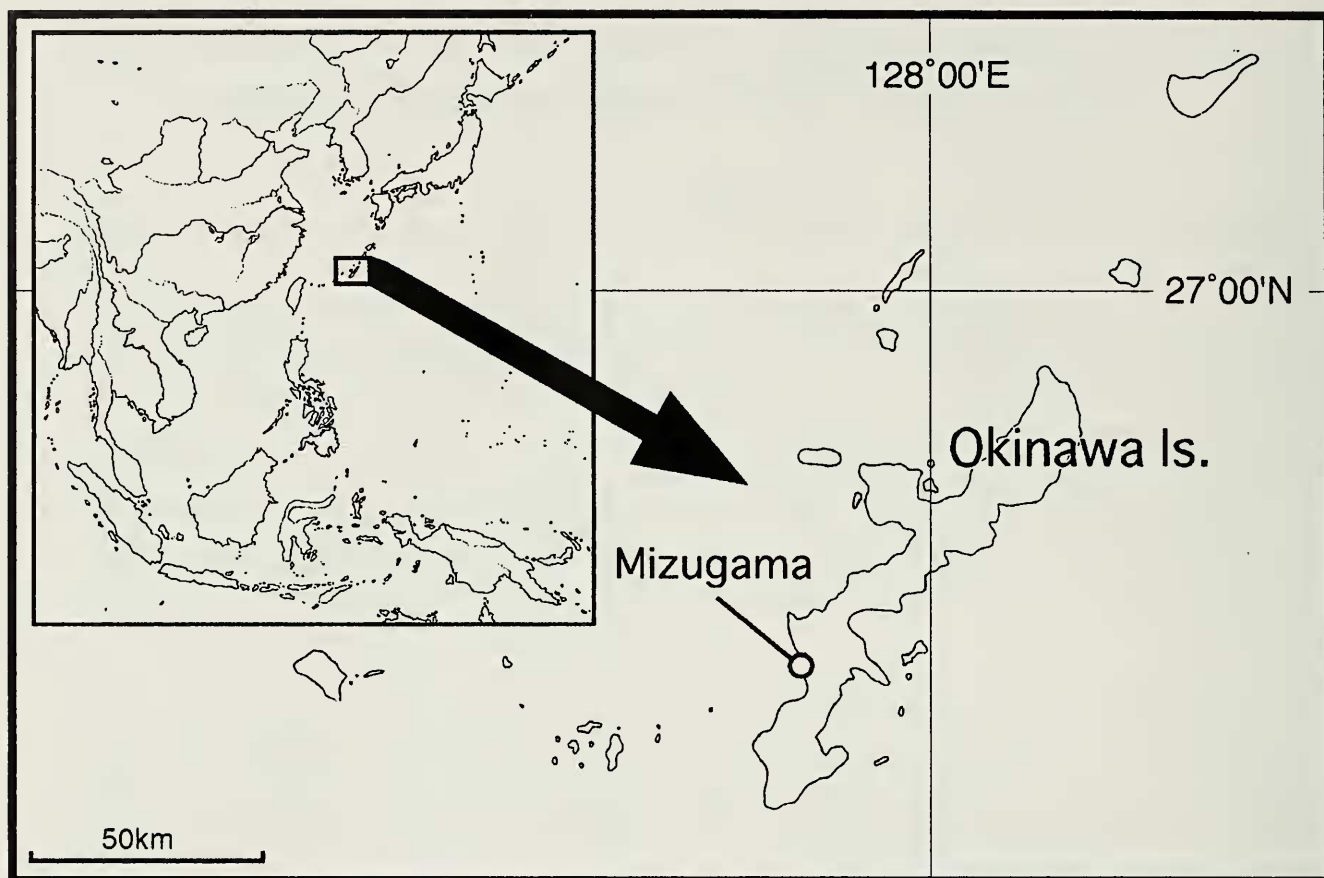


Fig. 1. Map of Okinawa Island and its vicinity, showing geographic location of Mizugama, the type locality of the species described in this study.

to yield numerous unknown entoprocts. Four new solitary entoprocts were collected from Okinawa Island and are described here.

Sampling of solitary entoprocts was carried out at a shallow reef flat in Mizugama ($26^{\circ}21'N$, $127^{\circ}45'E$), on the west coast of Okinawa Island, the Ryukyu Archipelago, Japan (Fig. 1). Glass slides were placed at a shady point at a depth of about 1 m and collected after two or four months. On one occasion, a glass bottle was collected. The epifauna on these objects was examined under a binocular stereomicroscope. Some of the entoprocts found from these objects were transferred to petri dishes and cultured in order to obtain the liberated buds or adult individuals that budded from the original individuals. Culturing, narcotization and fixation followed those of Iseto (2001). Drawings were prepared on the basis of fixed specimens, using a light microscope equipped with a camera lucida. Type spec-

imens are deposited in the National Science Museum, Tokyo (NSMT).

Family Loxosomatidae Hincks, 1880

Genus *Loxosomella* Mortensen, 1911

Loxosomella stomatophora, new species
Figs. 2, 3

Type material.—Holotype: adult (NSMT-Ka 52) settled on a glass slide submerged on the reef flat for two months at a depth of about 1 m in Mizugama, Okinawa Island, Japan, collected on 30 Aug 2001. Paratypes: 50 adults (NSMT-Ka 53) collected with the holotype. 19 adults (NSMT-Ka 54) settled on a glass slide submerged at the type locality for two months, collected on 25 Apr 2001.

Etymology.—Specific name *stomatophora* derived from Greek *stoma* = mouth + latinized greek suffix *phorus* = bearing, carrying, in allusion to the resemblance of the roundish body, in abfrontal view, to

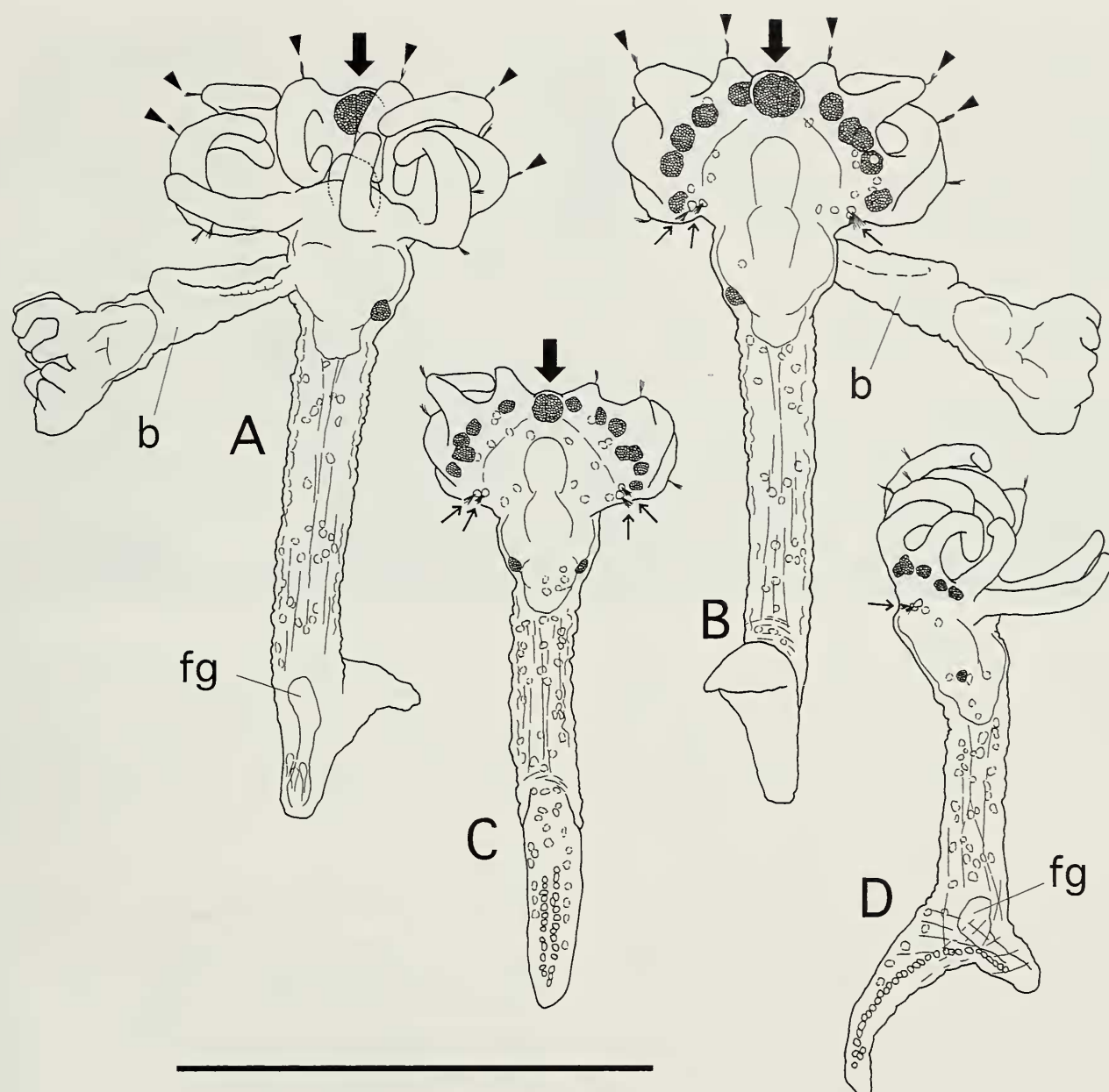


Fig. 2. Drawings of *Loxosomella stomatophora*, new species. A, B, holotype (NSMT-Ka 52) with large bud (b) in frontal and abfrontal views, respectively. C, D, paratype (NSMT-Ka 53) in abfrontal and lateral views, respectively. Thick arrows, characteristic roundish organ consisting of two large granular cells. Thin arrows, ciliary bundles at both lateral sides of the calyx. Arrowheads, ciliary bundles at outer side of tentacles. fg, foot gland. Scale bar = 500 μ m.

plant stomata; both are roundish structures consisting of two cells. Used as an adjective.

Description.—Total length (from basal attachment position of foot to upper-most level of tentacular membrane) 330 to 780 μ m. Tentacle number 10 in all specimens. Ciliary bundle present at outer side of each tentacle (arrowheads in Fig. 2). Two ciliary bundles present at each lateral side of calyx at level of upper side of stomach (thin arrows in Fig. 2; Fig. 3D). A peculiar round-

ish organ consisting of two large granular cells, 30 to 50 μ m in diameter, present at margin of tentacular crown between two abfrontal tentacles (thick arrows in Figs. 2, 3; Fig. 3B, C). Granular cells of 15 to 25 μ m in diameter present at margin of tentacular crown (thin arrows in Fig. 3A; Fig. 3E) and, in most specimens, at each lateral side of stomach. Stalk as long as or a little longer than calyx. Foot with foot gland and foot groove present both in bud and adult. Oral-most pair of tentacles bend outward

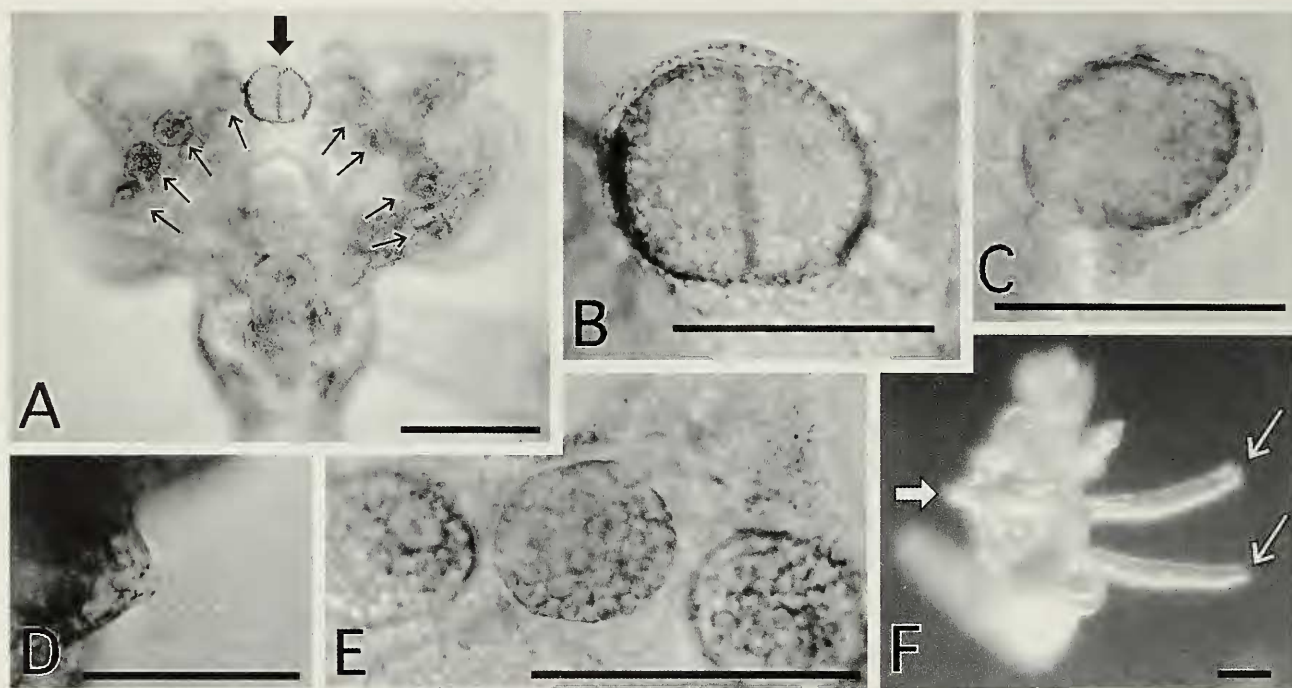


Fig. 3. Photographs of *Loxosomella stomatophora*, new species. A, abfrontal view of calyx of holotype (NSMT-Ka 52). Thick arrow indicates characteristic roundish organ. Granular cells at margin of calyx indicated with thin arrows. B, enlargement of roundish organ in A. C, left-side view of the roundish organ (paratype: NSMT-Ka 53). D, ciliary bundle at lateral side of calyx (paratype: NSMT-Ka 53). E, enlargement of granular cells at margin of calyx in A. F, upper view of living specimen. Note that frontal two tentacles (thin arrows) are extended frontally. The roundish organ indicated with thick arrow. Scale bars: A, F = 100 μ m; B–E = 50 μ m.

relative to other tentacles in expanded living specimens (Fig. 3F) but are often arranged as the other tentacles in fixed specimens.

Reproduction.—Buds emerge from the laterofrontal areas of the calyx at the level with the upper half of the stomach, attached by the foot tips. A maximum of four buds were observed simultaneously on a single parent. Sexual reproduction has not been observed.

Discussion.—The most characteristic feature of *Loxosomella stomatophora* lies in its possession of a peculiar roundish organ at the abfrontal margin of the tentacular crown. *Loxosomella vivipara* Nielsen, 1966 is also reported to have a specialized organ at the same position as *L. stomatophora* (Nielsen 1966, Emschermann 1993). The organ in *L. vivipara* is, however, found only in some individuals, and its form varies among individuals. In some individuals, it is a hollow cup-shaped structure with a plug of secretion and consists of 7

to 16 cells (Nielsen 1966), and in others it is a more conspicuous bell-shaped structure consisting of more than 25 cells (Emschermann 1993). The roundish organ of *L. stomatophora* is, however, composed of only two cells, showing only little variation among all the individuals examined in this study. *Loxosomella vivipara* is also distinct from *L. stomatophora* in having lateral wings on the calyx and conspicuous lateral pockets at the stomach (Nielsen 1966). Moreover, the tentacle number, which is invariably 10 in *L. stomatophora*, is 12 to 16 in *L. vivipara*.

Loxosomella lecythifera, new species
Figs. 4, 5

Type material.—Holotype: adult (NSMT-Ka 55) found on a glass bottle collected at the reef flat in Mizugama, Okinawa Island, Japan on 25 Oct 2000. Paratypes: Two adults (NSMT-Ka 56) collected with the holotype. 52 adults (NSMT-Ka

57), 473 adults (NSMT-Ka 58), and 41 adults (NSMT-Ka 59) settled on glass slides submerged on the type locality for two months, collected on 28 Mar 2001, 29 Jul 2001, and 29 Sep 2001, respectively. An adult (NSMT-Ka 60) having larvae in its calyx, collected with NSMT-Ka 58. A liberated bud (NSMT-Ka 61) budded from the holotype. A liberated bud (NSMT-Ka 62) budded from an adult collected with the holotype. Three liberated buds (NSMT-Ka 63) budded from adults settled on glass slides submerged on the type locality for two months, collected on 30 Jan 2001.

Etymology.—Specific name *lecythifera* from Latin *lecythus* = bottle or flask + suffix *-fer*, *-fera*, *-ferum* to bear or have, in allusion to the bottle-shaped appendages of the species. Used as an adjective.

Description.—Adult: Total length (from basal tip of stalk to uppermost part of tentacular membrane) up to 1650 μm . Tentacle number 14 to 22. Stalk one and a half to two times longer than calyx. Adult without foot. A pair of lateral sense organs present at level of upper side of stomach (thick arrows in Figs. 4A–C, 4E, 5A; Fig. 5C). Four to seven bottle-shaped appendages, 40 to 55 μm in length, present at abfrontal side of calyx at levels above the lateral sense organ (thick arrows in Figs. 4A–C, 4E, 5A; Fig. 5B, E). Many cylindrical appendages of 20 to 30 μm long present at calyx and stalk (Fig. 5D). Whole body, except tentacular crown, often covered with detritus. Liberated bud: Total length (from posterior tip of foot to anterior-most point of tentacular membrane between two abfrontal tentacles) 560 to 770 μm . Tentacle number 14 to 16. Stalk very short. Foot with foot gland and foot groove present. A pair of lateral sense organs present (thick arrows in Figs. 4D, 5F). Two to five bottle-shaped appendages comparable to those in adult specimens present (thin arrows in Figs. 4D, 5F).

Reproduction.—Buds emerge from the laterofrontal areas of the calyx at the level of the upper side of the stomach, attached by tips of their feet. A maximum of six

buds were observed simultaneously on a single parent. An embryo was found in the calyx of an individual (NSMT-Ka 60, Fig. 5G) collected in July. The size of the embryo is approximately 100 μm in diameter.

Discussion.—The distinguishing feature of *Loxosomella lecythifera* lies in its possession of several bottle-shaped appendages. Some *Loxosomella* species are reported to have appendages of more irregular shape, but bottle-shaped appendages have not previously been reported from the congeners. *Loxosomella hispidula* Marcus & Marcus, 1968 resembles *L. lecythifera* in having appendages at the calyx and stalk, having a maximum of six buds, and having more than 20 tentacles (Marcus & Marcus 1968). However, the large appendages referred to as sense organs, at the middle level of the stomach in *L. hispidula* (Marcus & Marcus 1968; fig. 11) do not resemble any appendage of *L. lecythifera*.

Loxosomella intragemmata, new species
Figs. 6, 7

Type material.—Holotype: adult (NSMT-Ka 64) settled on a glass slide submerged on the reef flat for two months at a depth of about 1 m in Mizugama, Okinawa Island, Japan, collected on 29 Dec 2000. Paratypes: Seven adults (NSMT-Ka 65) collected with the holotype; 81 adults (NSMT-Ka 66), 389 adults (NSMT-Ka 67), and 25 adults (NSMT-Ka 68) settled on glass slides submerged on the type locality for two months, collected on 28 Feb 2001, 29 Jul 2001, and 29 Nov 2001, respectively. Five adults (NSMT-Ka 69) having larvae in their calyx, collected with NSMT-Ka 67. Three liberated buds (NSMT-Ka 70) budded from adults collected with the holotype.

Etymology.—Specific name *intragemmata* from Latin *intra* = within + *gemmatus*, with buds, from *gemma* = bud, in allusion to the bottle-shaped appendages of the species. Used as an adjective.

Description.—Adult: Total length (from basal tip of stalk to uppermost part of ten-

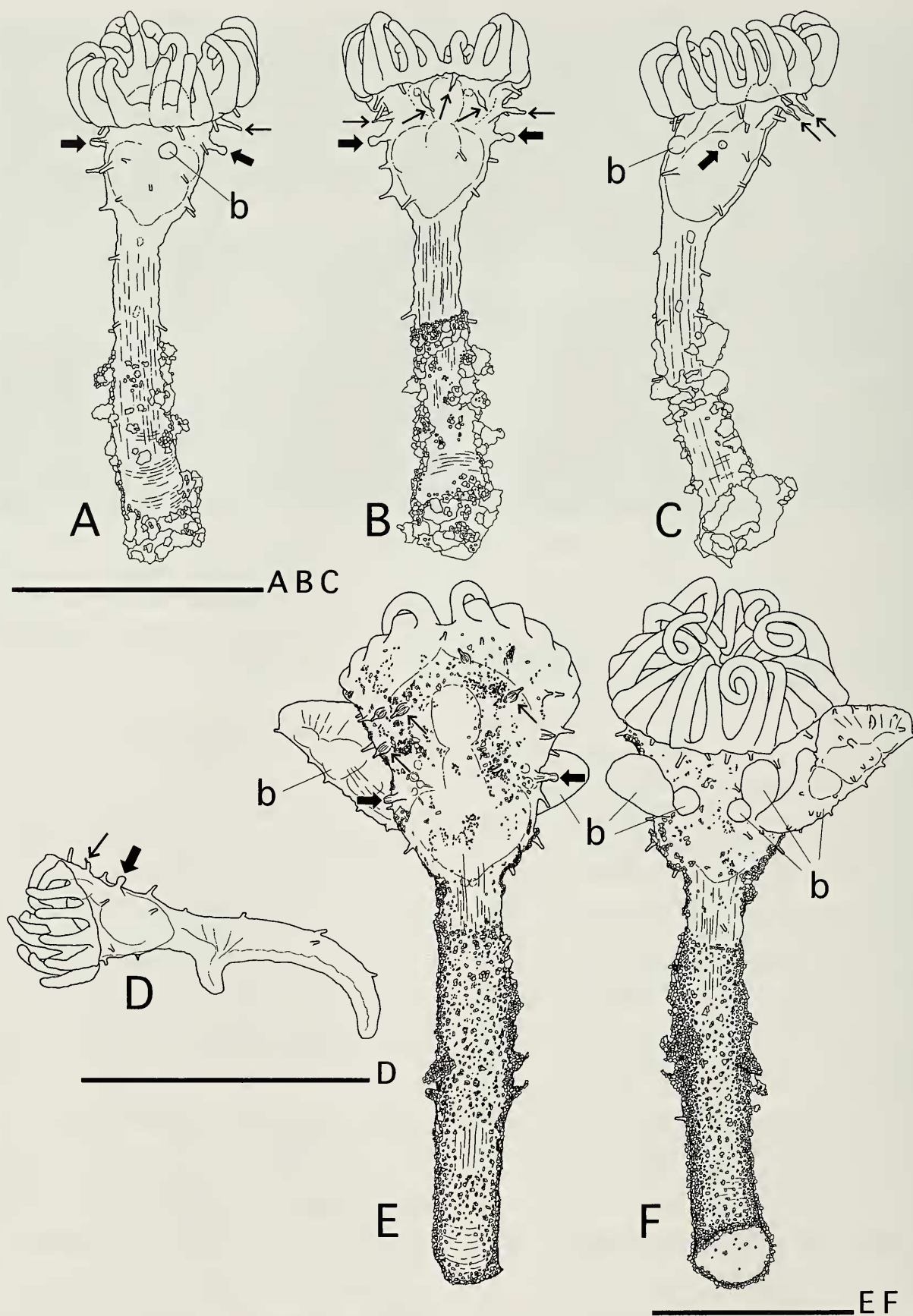


Fig. 4. Drawings of *Loxosomella lecythifera*, new species. A–C, adult holotype (NSMT-Ka 55) with small bud (b) in frontal, abfrontal, and lateral views, respectively. D, lateral view of liberated bud (paratype, NSMT-Ka 61) budded from holotype. E, F, large adult specimen (paratype, NSMT-Ka 57) with five buds (b) in abfrontal and frontal views, respectively. Thick arrows, lateral sense organs. Thin arrows, some of bottle-shaped appendages. Scale bars = 500 μ m.

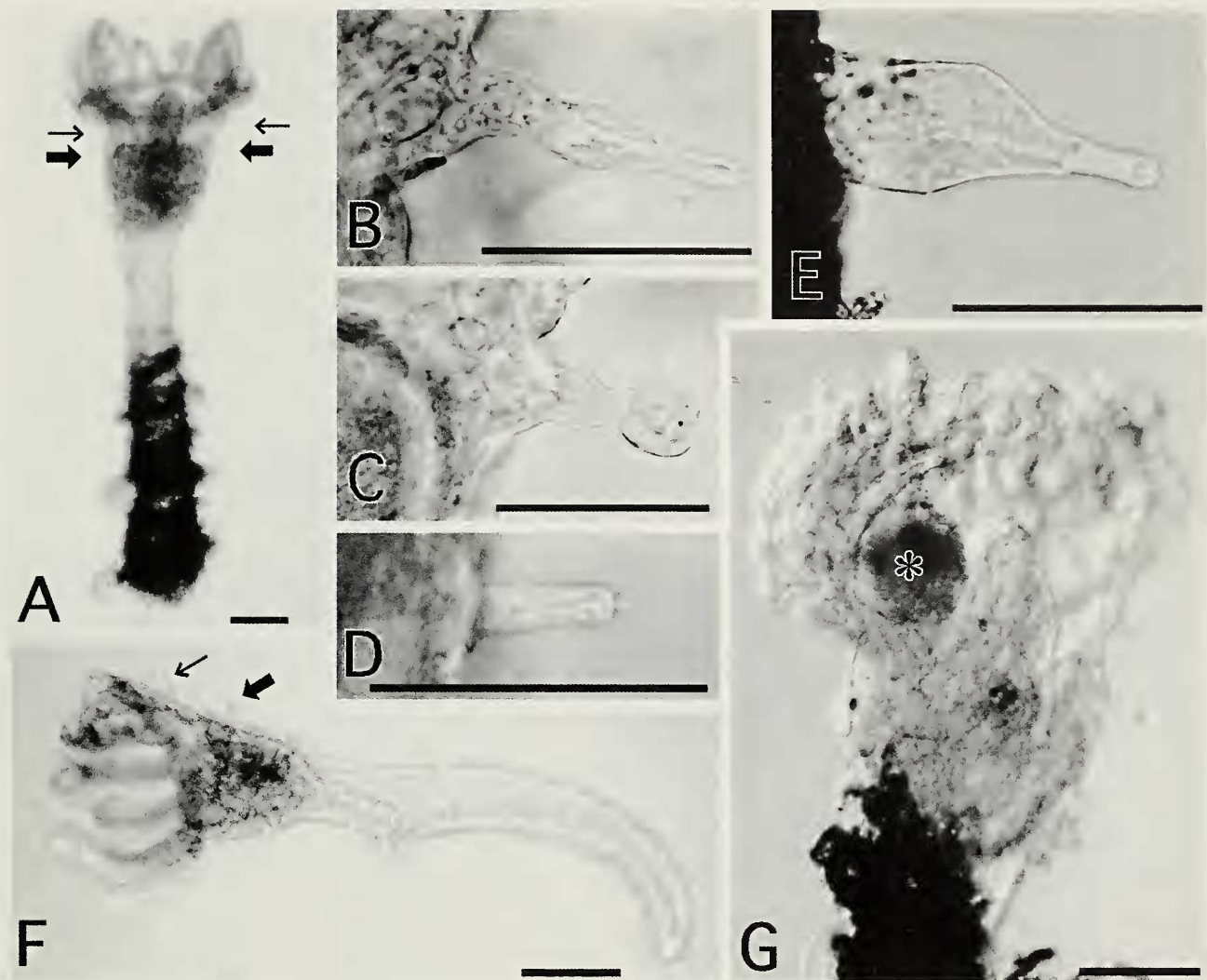


Fig. 5. Photographs of *Loxosomella lecythifera*, new species. A–D, adult holotype (NSMT-Ka 55). A, abfrontal view of whole body. B, characteristic bottle-shaped appendage. C, lateral sense organ. D, cylindrical appendage at calyx. E, bottle-shaped appendage of large paratype specimen (NSMT-Ka 57). F, left side view of liberated bud (paratype, NSMT-Ka 61). G, latero-abfrontal view of calyx of paratype (NSMT-Ka 60) brooding larva (asterisk) in its calyx. Thick arrows, lateral sense organs. Thin arrows, bottle-shaped appendages. Scale bars: A, F, G = 100 μ m in; B–E = 50 μ m.

tacular membrane) up to 1260 μ m. Tentacle number 10 to 15. Stalk two to three times longer than calyx. Adult without foot. A pair of lateral sense organs present (arrows in Fig. 6). Liberated bud: Total length (from posterior tip of foot to anterior-most point of tentacular membrane between two abfrontal tentacles) 420 to 480 μ m. Tentacle number 10. Foot with foot gland and foot groove present. A pair of lateral sense organs present (arrows in Figs. 6D, 7C).

Reproduction.—Buds emerge internally at the laterofrontal areas of the calyx at the level with the upper half of the stomach. When the spherical small buds become

elongate, the buds begin to protrude from the parent's body (Fig. 7B). Buds attached by the tips of their feet. A maximum of three buds were observed simultaneously on a single parent. Embryos (up to four) were found in the calyx of five individuals (NSMT-Ka 69, Fig. 7D) collected in July. The size of the embryos are up to 120 μ m.

Discussion.—The most remarkable feature of *Loxosomella intragemmata* is that the buds are formed internally in contrast to most other solitary species in which the buds emerge from the outer surface of the parent. Similar internal budding was reported for *L. bocki* Franzén, 1966 found on

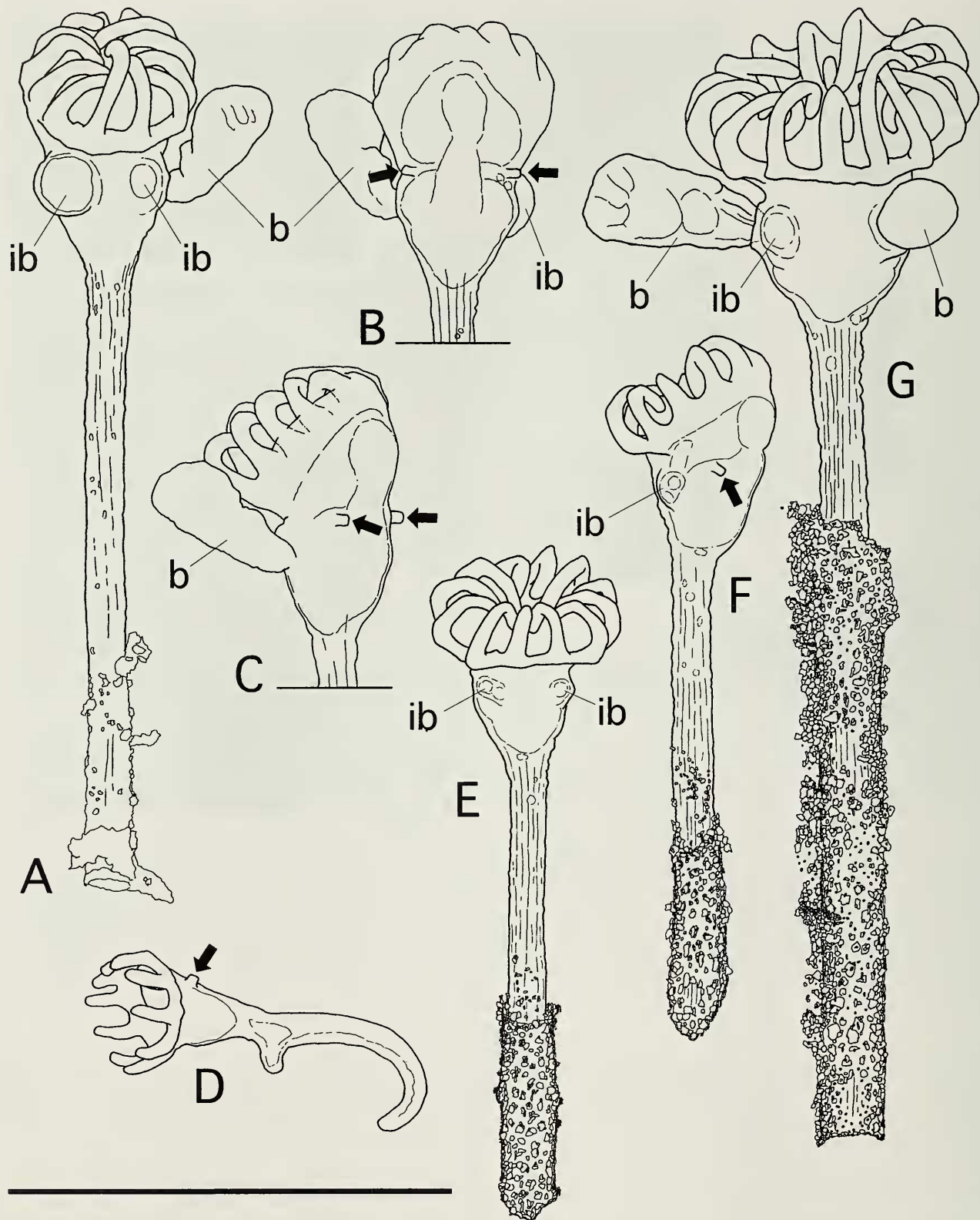


Fig. 6. Drawings of *Loxosomella intragemmata*, new species. A–C, adult holotype (NSMT-Ka 64) with two internal buds and bud protruding from its pocket at calyx, in frontal, abfrontal, and lateral views, respectively. D, left side view of liberated bud (paratype, NSMT-Ka 70). E, F, small adult specimen (paratype, NSMT-Ka 67) with internal buds in early stage, in frontal and lateral views, respectively. G, large paratype specimen in frontal view. Arrows, lateral sense organs. b, protruding bud. ib, internal bud. Scale bar = 500 μ m.

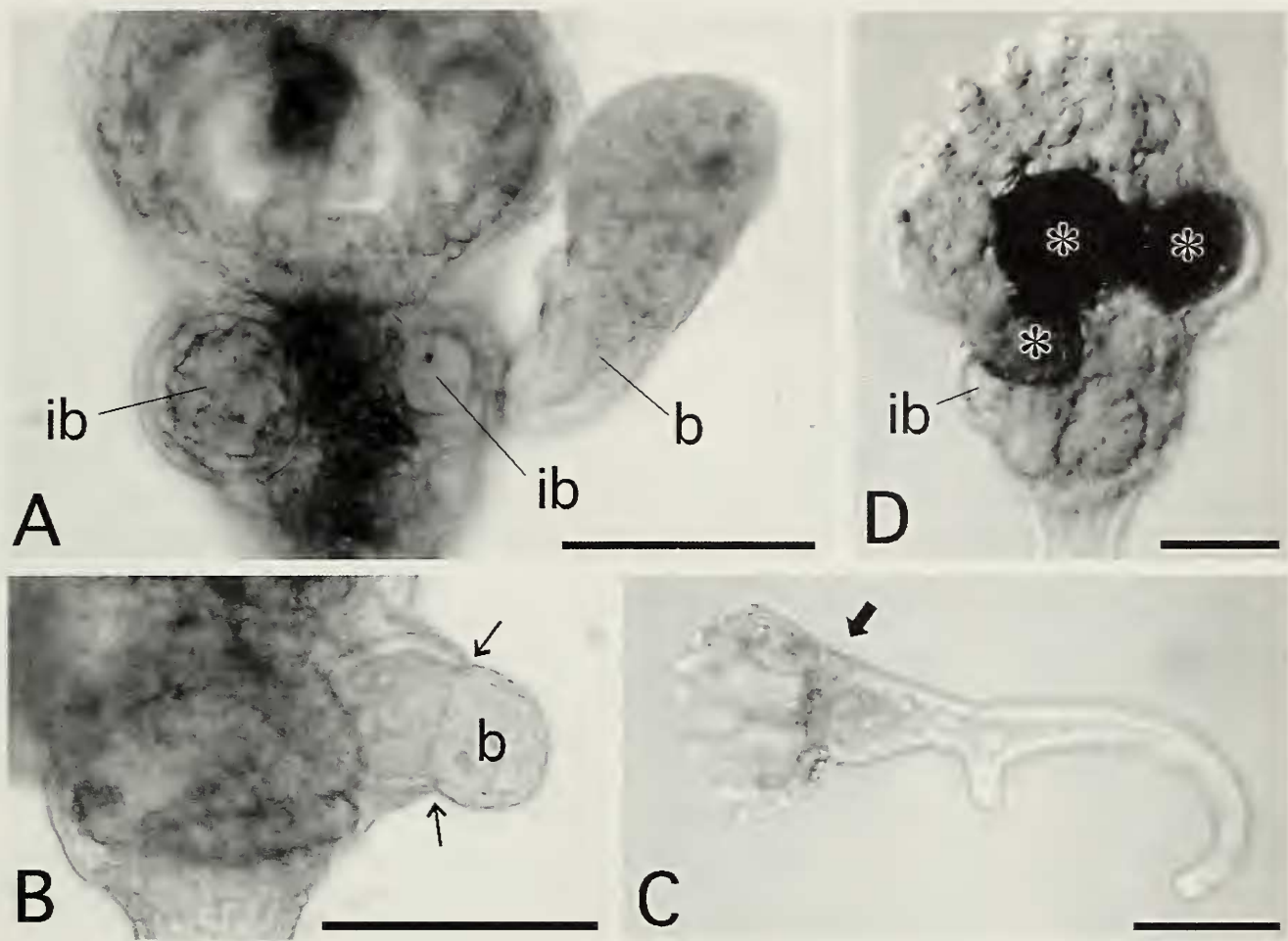


Fig. 7. Photographs of *Loxosomella intragemmata*, new species. A, frontal view of calyx of holotype (NSMT-Ka 64) with two internal buds (ib) and protruding bud (b). B, frontal view of lower half of calyx of paratype (NSMT-Ka 65) showing bud which has just begun to protrude from parent body (b). Arrows indicate opening of pocket from which bud protrudes. C, left side view of liberated bud (paratype, NSMT-Ka 70). Arrow indicates lateral sense organ. D, latero-abfrontal view of calyx of paratype specimen (NSMT-Ka 69) with internal bud (ib) and three larvae (asterisks). Scale bars = 100 μ m.

a sponge collected at the Gilbert Islands. *Loxosomella bocki* is, however, distinct from *L. intragemmata* in retaining the foot in the adult stage, having a short (slightly shorter than the calyx) stalk, and lacking the lateral sense organ at the calyx (Franzén 1966). The internal buds, in both species, may occur from deep epitherial pockets, a pair of depressions at the laterofrontal area of the calyx (Franzén 1966). Budding from less deep pockets was reported from some congeneric species such as *L. raja* (Schmidt, 1876), *L. pes* (Schmidt, 1876), *L. antis* Krylova, 1985, and *L. cricketae* Nielsen, 1966. However, these pockets have outer openings unlike those of the above

two species, which appear completely covered by the epidermis.

Genus *Loxocorone* Iseto, 2002

Loxocorone dicotyledonis, new species
Figs. 8, 9

Type material.—Holotype: adult (NSMT-Ka 71) settled on a glass slide submerged on the reef flat for four months at a depth of about 1 m in Mizugama, Okinawa Island, Japan, collected on 5 Jul 2000. Paratypes: 138 adults (NSMT-Ka 72) derived by budding from three adults found on a slide submerged at the type locality for two months, collected on 11 Apr 1999. An

adult (NSMT-Ka 73) derived by budding with NSMT-Ka 72.

Etymology.—Specific name *dicotyledonis*, Latinized Greek *di* = two + *cotyledon* = seed leaf of a plant + suffix *-is*, = resemblance, in allusion to the laterally elongated tentacular crown resembling the seedling of a dicotyledonous plant. Used as an adjective.

Description.—Total length (from basal attachment position of foot to upper-most level of tentacular membrane) 380 to 900 μm . Number of tentacles 18 to 28. Tentacles alternately directed laterally and frontally. Both lateral sides of the tentacular crown expand laterally making the tentacular crown bilobed rather than circular. Stalk as long as calyx. Foot with foot gland and foot groove present both in bud and adult. Foot gland long (about 300 μm), reaching to the middle level of the stalk. Lateral sense organs absent. Glandular cells of about 20 μm in diameter scattered in calyx (arrowheads in Fig. 9C; Fig. 9D, E), tentacles, stalk (arrows in Fig. 9F; Fig. 9G), and foot (Fig. 9H). These glandular cells often have external secretion (arrows in Fig. 9E, G, H).

Reproduction.—Buds emerge from the frontal areas of the calyx at the level of the upper side of the stomach, attached to their parent's calyx by the back of the upper part of the stalk. A maximum of two buds were observed simultaneously on a single parent. Nevertheless, only one bud grew large at a time. Large buds have the same proportion as the adults but are smaller and have a lower tentacle number than the adults. When the bud has grown large enough to detach, the parent bends its stalk frontally. Then, the foot of the bud attaches to the substratum and the bud detaches from the parent. *Loxocorone dicotyledonis* showed active asexual propagation under laboratory culture fed with microalgae. The 138 paratype specimens (NSMT-Ka 72) were derived from only three individuals after 40-days culture. Sexual reproduction has not been observed.

Discussion.—The bilobed shape of the tentacular crown is the distinguishing feature of *Loxocorone dicotyledonis*. The deviation of the shape of the tentacular crown from the normal circular shape is also observed in some other loxosomatids (e.g., *Loxosoma nung* Nielsen, 1996). In those species, however, the deviation is not as conspicuous as in *Loxocorone dicotyledonis*. The characteristic arrangement of the tentacles that are alternately directed laterally and frontally was also reported in *Loxocorone allax* Iseto, 2002. However, the two species are easily distinguishable because in *L. allax* the foot degenerates in the adult stage unlike *L. dicotyledonis* in which all individuals retain the foot throughout the life. The shape of the bud is also considerably different in the two species. Unlike the bud of the present species that has a definite stalk and foot-shaped foot, that of *L. allax* lacks stalk and has a long, cylindrical foot the length of which reaches one and half times as long as its calyx.

Another noteworthy feature of *Loxocorone dicotyledonis* lies in its possession of the glandular cells with external secretion at abfrontal side of the calyx, tentacle, stalk, and foot. Among these glandular cells, two distinct cell types seem to be distinguishable. The cells at calyx and tentacles are spherical, secreting vague mucous material (arrows in Fig. 9E), whereas the cells at stalk and foot are more irregular, secreting more clearly outlined substance (arrows in Fig. 9G, H).

General Discussion

Most of the solitary entoprocts hitherto described were found on host animals such as polychaetes, bryozoans, sipunculans and sponges (summarized in Nielsen 1964, Soule & Soule 1965). All of the ten species recorded in the Ryukyu Archipelago, however, had settled on glass slides or a glass bottle, and thus, do not depend on host animals (Iseto 2001, 2002, present study). Some of these species are also found on

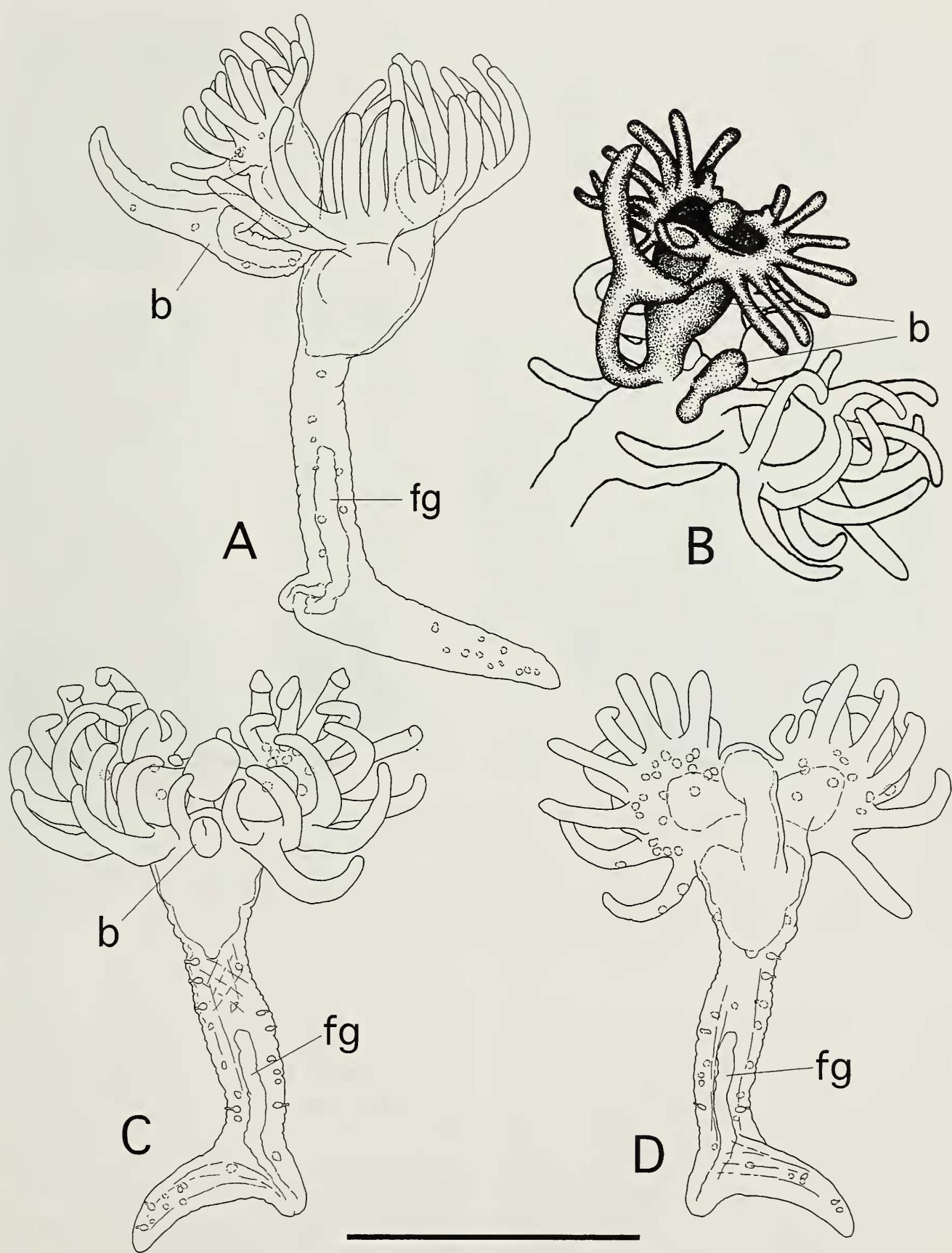


Fig. 8. Drawings of *Loxocorone dicotyledonis*, new species. A, lateral view of holotype (NSMT-Ka 71) with a large bud. B, calyx of a paratype (NSMT-Ka 72) with a large and a small bud (shaded). C, D, paratype (NSMT-Ka 73) with a small bud, in frontal and abfrontal views, respectively. b, bud. fg, foot gland. Scale bar = 500 μ m.

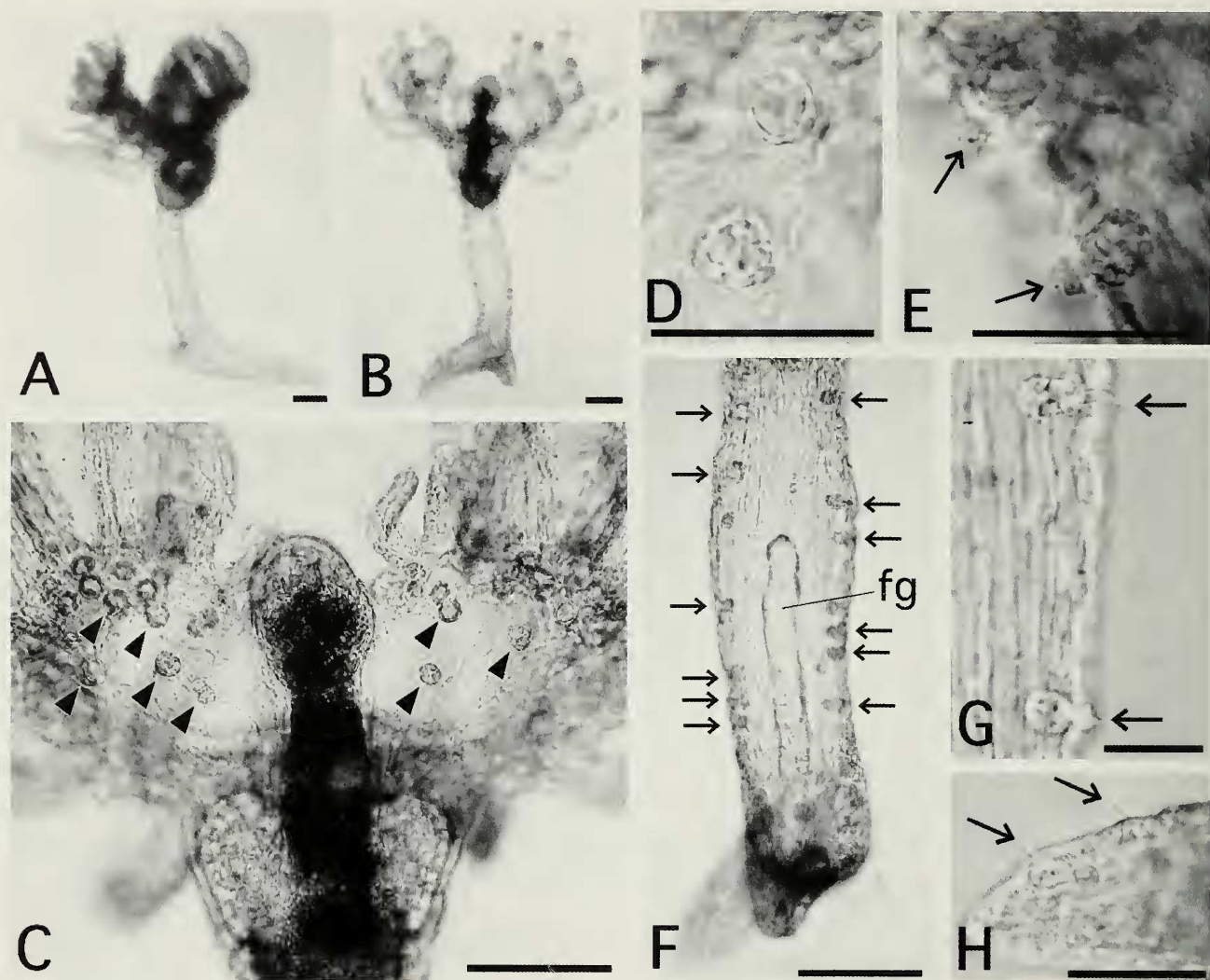


Fig. 9. Photographs of *Loxocorone dicotyledonis*, new species. A, lateral view of holotype (NSMT-Ka 64) with large bud. B–H, paratype (NSMT-Ka 72). B, frontal view. C, abfrontal view of the calyx. Arrowheads indicate glandular cells. D, E, enlargement of glandular cells at calyx in surface view and lateral views, respectively. Arrows indicate external secretion. F, frontal view of lower half of animal. Arrows indicate glandular cells at lateral sides of stalk. fg, foot gland. G, enlargement of glandular cells at stalk. H, posterior tip of foot. Arrows in G and H indicate external secretion of glandular cells. Scale bars: A–C, F = 100 μ m; D, E, G, H = 50 μ m.

dead coral fragments and shell remains collected from reef flats, and thus, non-living substrata are undoubtedly their natural habitat. Although commensal species may possibly be discovered from this area in future studies, non-living substrata seem to be a common habitat for the solitary entoprocts of this area. The studies in the Ryukyu Archipelago aroused the question of whether solitary entoprocts are an animal group that is commensal in general, as noted in many publications (Hyman 1951, Ruppert & Barnes 1994, Nielsen 2001). As well as the ten species from the Ryukyu Archipelago, *Loxosomella olei* du Bois-Reymond-Mar-

cus, 1957 and *Loxosoma isolata* Salvini-Plawen, 1968 were found only on non-living substrata. Furthermore, some species were found on several animals of different phyla and also on non-living substrata (cf. Nielsen 1964), suggesting that a host animal is not essential for these entoprocts. Nielsen (1964) insisted that the solitary entoprocts are “energy commensals” that depend on the host-made water current to obtain food particles. The basis of his argument was that commensal species are mostly situated at points where water currents are made by the hosts. However, the ten species found from the Ryukyus live

without host animals. Despite this, they do not seem to have any adapted characteristics to make stronger water currents than the commensal species. This indicates that most commensal species can also produce sufficient water currents by themselves. Non-commensal species were often found at small cleavage points of stones, which may offer protection from larger animals; the densely aligned glass slides used in my studies may have provided a similar condition. My observation suggests that the habitat preference of solitary entoprocts is to avoid predators. The body surface of the host animals or inner side of host tubes also help solitary entoprocts to avoid predators, although it is still probable that short-tentacled species depend on the host-produced water current.

To my knowledge, there is no report on long-term laboratory culture of solitary entoprocts. In this study, however, vigorous budding in several generations was induced on *Loxocorone dicotyledonis* cultured in petri dishes (ca. 50 ml). They were fed surplus amount of microalgae, *Nannochloropsis* (Marine Chlorella 100, Marine-bio Co., Japan), and the seawater was exchanged after about 30 minutes. The microalgae and excrements that accumulated under the dishes were cleaned up several times a week using a thin wood stick. I did not try long-term culture for the other three species described in this study, but the buds that were already present at the collection time were grown and released within a week. Among the six other species that I previously reported from the same region, *Loxomitra mizugamaensis* Iseto, 2002, *L. tetrorganon* Iseto, 2002 and *Loxocorone alax* Iseto, 2002 proliferated similarly to *Loxocorone dicotyledonis* by the same culture method. For *Loxosomella monocera* Iseto, 2001, the culture was successful, but the budding and growth rate was very slow, suggesting that the culture method was not adequate for this species. For *L. aloxiata* Iseto, 2001, budding could be induced but the newly liberated buds never successfully

fixed on substrata and failed to reach the normal adult form. At present, it is not understood why some species are tolerant to the culture condition and others are not. *Loxosomella lappa* Iseto, 2001 was found attached to a glass plate that had been kept in a bucket with seawater and coral rubble on which the original individual(s) were attached. In this case, diet was not supplied, but the animals proliferated with contaminated detritus. The seawater was not exchanged and the bucket was left for about one month with air supply. This is the second culture method I have tried and also an easy collecting method for inconspicuous species that are difficult to find directly from the original, complicated substrata.

In my surveys in the Ryukyu Archipelago, most of the non-commensal entoprocts were found on glass slides used as settlement panels. Ryland & Austin (1960) found two solitary species, *Loxomitra kefersteinii* (Claparède, 1867) and *Loxosomella antedonis* Mortensen, 1911 on plastic settlement panels. Settling panels seem to be an effective way to collect non-commensal solitary entoprocts. Adaptation of this method may reveal a rich entoproct fauna without host animals in areas outside the Ryukyu Archipelago, including areas from which only commensal species have been reported in the past studies.

Acknowledgments

I wish to express my gratitude to Professor Shunsuke F. Mawatari (Hokkaido University) and Dr. Euichi Hirose (University of the Ryukyus), for their valuable advice and encouragement. Thanks are also due to Dr. Masatsugu Yokota (University of the Ryukyus) for helpful suggestions on nomenclature. This study is supported in part by a subvention to graduate students by Research Institute of Marine Invertebrates and the Sasagawa Scientific Research Grant from the Japan Science Society.

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